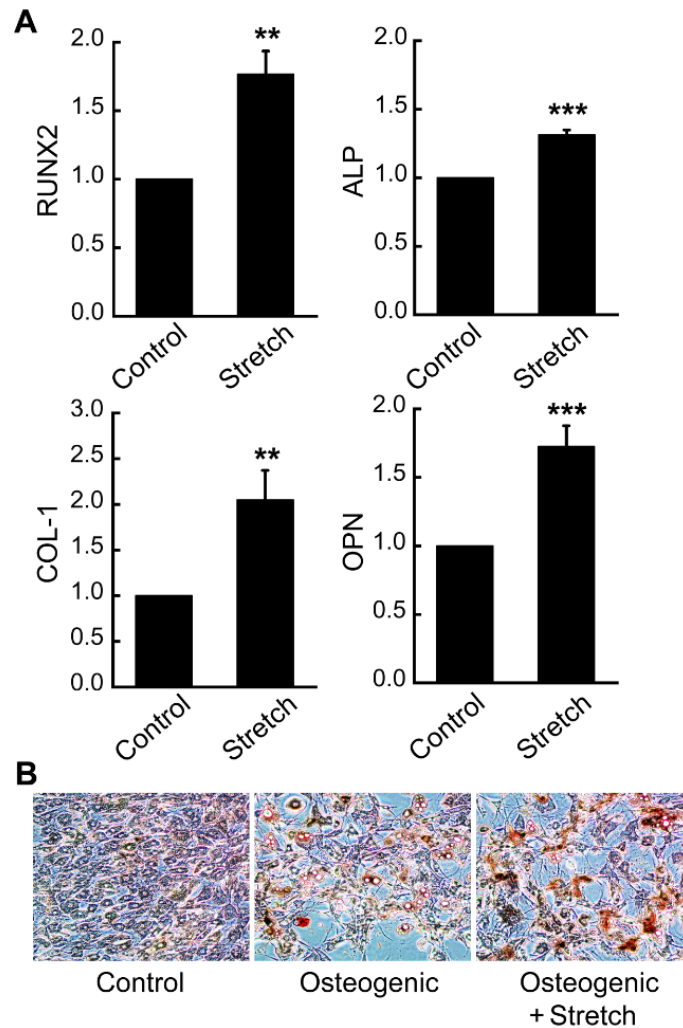


## YAP mechanotransduction under cyclic mechanical stretch loading for mesenchymal stem cell osteogenesis is regulated by ROCK

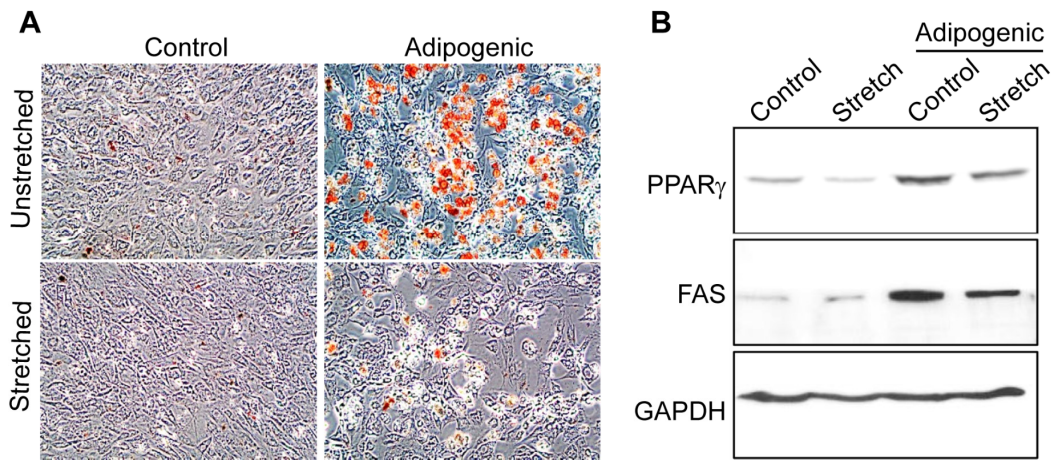
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### Supplementary Material

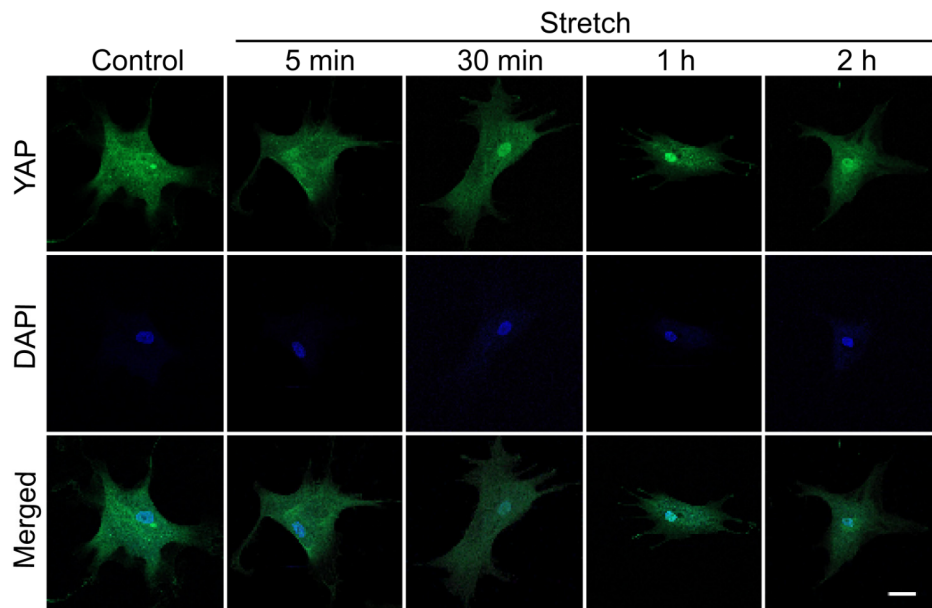


### Supplementary Fig. S1. Cyclic stretch promotes the osteogenic differentiation of MSCs.

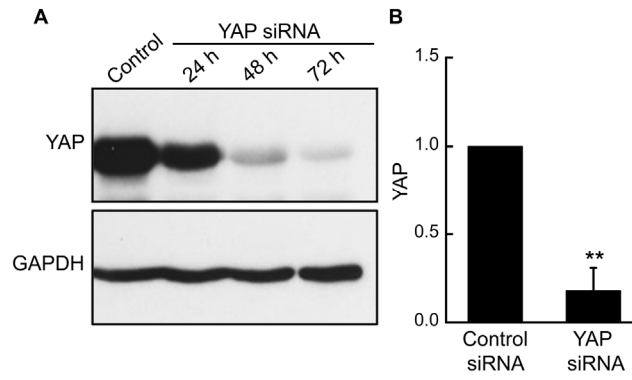
MSCs were stretched for 3 days (1 h/day) with the onset of the exposure to osteogenic induction media. (A) Osteogenic gene expressions by qRT-PCR on day 7 (n = 6). (B) Alizarin red bone mineral staining on day 21. \*\*:  $p < 0.01$  and \*\*\*:  $p < 0.001$  compared with control.



**Supplementary Fig. S2. Cyclic stretch suppresses the adipogenic differentiation of MSCs.** MSCs were seeded on the stretch plate for 2 days until they reached confluence. Adipogenesis was induced by exposure to 1  $\mu\text{g/ml}$  insulin, 0.5 mM isobutyl-methylxanthine, and 10  $\mu\text{M}$  dexamethasone for 2 days, and stretch was applied for 1 h/day. After that, adipogenic maintenance medium (1  $\mu\text{g/ml}$  insulin) was given for additional 8 days. (A) Lipid accumulation shown by oil red O staining. (B) Western immunoblotting of adipogenic markers, PPAR $\gamma$  and FAS, after 2 days of adipogenic induction without or with stretch.



**Supplementary Fig. S3. Human MSCs show cyclic stretch-induced YAP nuclear transport.** hMSCs (Lonza, PT-2501, 25 year-old female) were exposed to cyclic stretch loading at 10% strain and 1 Hz frequency. Immunofluorescent imaging of YAP (green) and DAPI (blue). Scale bar = 20  $\mu\text{m}$ .



**Supplementary Fig. S4. YAP silencing via siRNA transfection.** (A) MSCs were transfected with control siRNA or YAP siRNA, and YAP expression was assessed by western blotting. (B) At 48 h post-transfection, YAP expression was significantly decreased (n = 3). \*\*: p < 0.01.